

Interseeding Novel Cool-Season Annual Legumes to Improve Bermudagrass Paddocks

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ABSTRACT

Interseeding nontraditional, cool-season legumes into bermudagrass [*Cynodon dactylon* (L.) Pers.] paddocks was evaluated as an approach to increasing the quality and duration of forage production and replacing a portion of the N fertilizer required in the southern Great Plains. We compared the effects of interseeding either grass pea (*Lathyrus sativa* L. 'AC-Greenfix') or lentil (*Lens culinaris* Med. 'Indianhead') with N fertilizer rates of 0, 45, or 90 kg ha⁻¹ N. All plots received 60 kg P₂O₅ ha⁻¹ in early March. The legume and fertilizer treatments were imposed in mid-March during 2001, 2002, and 2003. Forage samples were clipped from 0.25 m⁻² quadrats on five sampling dates between 1 May and 15 July each year. Yield, N concentration, species composition, and in vitro digestible dry matter (IVDDM) were determined. Year, sampling date, and treatment showed significant ($P < 0.05$) effects, as did the two-way interactions between all three factors. Total end-of-season standing dry matter of bermudagrass and grass pea was 5550 ± 423 (SEM) kg ha⁻¹, which was similar to biomass production with 45 kg ha⁻¹ N (5305 ± 570 kg ha⁻¹) and less than that produced with 90 kg ha⁻¹ N (7785 ± 725 kg ha⁻¹). Forage N and IVDDM concentrations for the grass pea treatment were 34 and 6% higher than for bermudagrass, but N and IVDDM concentrations of the forage mixture were intermediate between the higher N rates. Although additional studies are needed to optimize management for the interseeded legumes, we conclude that this practice can improve the quality and duration of bermudagrass forage production in this region.

THE RISING COST of commercial fertilizer has renewed interest in grass-legume forage systems. Baylor (1974) noted that including legumes in paddocks of perennial warm- or cool-season grasses usually resulted in increased yields, better forage quality, and improved seasonal distribution of forage production. Grass-legume mixtures also may increase animal performance and carrying capacity (Seguin, 1998). Legumes generally have a greater relative feed value than warm-season grasses (Ball et al., 2002), can improve the protein content of the pasture, and provide a renewable source of N for plant growth, which decreases N fertilizer requirements (Hoveland, 1989).

Several studies have demonstrated the value of planting legumes in established paddocks of warm-season perennial grasses (Templeton and Taylor, 1975; Jung et al., 1985; Brown and Byrd, 1990). However, Marten (1985) reported that cool-season legumes might not be satisfactory companion species for warm-season grasses due to differences in seedling vigor, optimum time for establishment and growth, and low persistence. Also, spring growth of a warm-season grass stand may

be inhibited by competition for water, nutrients, and sunlight from a cool-season legume.

Although interseeding legumes into established grass stands is a common practice in some regions, little information is available regarding forage yield, quality, and compatibility of short-lived cool-season legumes planted into stands of warm-season perennial grasses in the southern Great Plains. The objective of this study was to compare forage yield and nutritive value of pure stands of bermudagrass with stands of bermudagrass interseeded with two cool-season annual legumes (grass pea and lentil) that are novel to the southern Great Plains.

MATERIALS AND METHODS

Study Site

This study was conducted from March through June of 2001 to 2003 at the USDA-ARS Grazinglands Research Laboratory near El Reno, OK (35°40' N, 98°0' W, elevation 414 m). The experimental site was situated in a 3.2-ha paddock of 'Midland' bermudagrass established in 1991 from sprigs. Soils were a Brewer silty clay loam (fine-loamy, mixed, thermic, Udic Rhodustalfs; USDA-NRCS, 1999). The average daily maximum and minimum temperatures for the experimental period was 25 and 16°C, respectively (Brock et al., 1995). The paddock was burned in late February of each year to remove the previous year's growth.

Five 3-by-20-m plots were established in each of three blocks, and all plots were fertilized with 60 kg P₂O₅ ha⁻¹ during 1 to 7 March of each year. Nitrogen fertilizer and interseeding treatments were randomly assigned to plots within blocks. Treatments included (i) interseeding with AC-Greenfix grass pea or (ii) Indianhead black lentil, (iii) fertilizing with urea at 45 kg ha⁻¹ N, (iv) urea at 90 kg ha⁻¹ N, or (v) no N fertilizer or legume. Nitrogen fertilizer was broadcast in mid-March of each year. Seeds of both legumes were treated with a commercial liquid inoculum containing *Rhizobium leguminosarum* (Liphatech, Milwaukee, WI)¹ and planted in 60-cm rows at 60 kg ha⁻¹ (75% germination) for grass pea and 20 kg ha⁻¹ (80% germination) for lentil on 15 March. No N fertilizer was applied to plots seeded with legumes. Seeding rate and row spacing provided 10 to 15 plants m⁻¹ row length. Treatments were repeated on the same plots each year.

Data Collection

Beginning 45 d after the legumes were seeded, whole-plant samples of legumes and grass were collected on five dates, four at approximately 9-d intervals [45–75 d after seeding (DAS)]

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Published in Crop Sci. 47:168–173 (2007).

Forage & Grazinglands
doi:10.2135/cropsci2006.02.0088

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Abbreviations: DAS, days after seeding; IVDDM, in vitro digestible dry matter.

Table 1. Precipitation recorded at the study site during the March to July growing season in 2001 through 2003 and the 25-yr seasonal average (1978–2003).

Month	Growing seasons			25-yr average
	2001	2002	2003	
	mm			
March	26	37	33	72
April	14	99	34	73
May	199	47	66	162
June	34	60	91	125
July	30	23	18	55
Total	303	266	242	487

and the fifth on 95 DAS. On each sampling date, three randomly selected 0.25-m² quadrates were clipped to a height of 2.5 cm within each plot. Samples were collected at new locations on each sampling date. Samples were dried in a forced-air oven at 55°C until a consistent weight was obtained, and weighed to calculate aboveground standing crop. Samples were separated into grass (grass standing crop) and legume (legume standing crop) fractions to partition the total yield. Samples were then ground to pass a 1.0-mm screen and analyzed for N concentration by complete combustion N analyzer (Leco CHN-1000, LECO Corp., St Joseph, MI). The IVDDM was determined by the two-stage technique of Tilley and Terry (1963) as modified by Monson et al. (1969).

Statistical Analysis

Data were analyzed by longitudinal data analysis (e.g., repeated measures) within mixed model procedures (Patetta, 2005). Treatments, DAS, and years were organized as fixed effects within individual experimental plots, which served as local subjects in analyses. The spatial power covariance structure was used to describe DAS effects in model development due to the uneven spacing of sampling dates and form of correlation among DAS. Statistical tests were restricted to main effects and two-way interactions, as the data sets lacked sufficient degrees of freedom ($n = 225$ for total and grass variables, $n = 90$ for legume variables) to test higher-order interactions or more complex DAS effects. Attempts at including more complex effects in the models developed by longitudinal analyses, or treating years in cross-sectional analyses (Patetta, 2005) did not produce stable variance/covariance matrices or functional models. Years were left as fixed effects, despite repeated application of treatments to plots. This approach was used because existing subroutines within PROC MIXED model analyses do not allow the analysis of double repeated measures (Patetta, 2005). The LSMEANS procedure (Littell et al., 1996) was used to test for differences in main and interaction effects, using $P = 0.05$ as the level of significance.

RESULTS AND DISCUSSIONS

The study was conducted over a series of years with poor growing conditions. Precipitation during the March through July growing season in 2001, 2002, and 2003 was only 62, 55, and 50%, respectively, of the 25-yr average of 487 mm (Table 1). Significant ($P < 0.05$) main effects related to DAS, treatments, and years were noted for yield, N, and IVDDM concentrations of grass, legume, and total standing crops (Table 2). The DAS \times treatment interactions were significant for all measured variables, as were DAS by year interactions for yield and IVDDM of grass forage, legume yield, and N content and yield of total forage. On the basis of the consistency of year main effects noted for most of the variables (except IVDDM of total forage), we concluded that climatic conditions within individual growing seasons had some impact on treatment responses.

Standing Crop

Yield from grass, legume, and total standing crops varied among years (Table 3). The low precipitation during the first 2 mo of the 2001 (Table 1) growing season appeared to cause a DAS \times year interaction in grass standing crop by limiting early production in 2001, while significant rains in May 2001 allowed the highest standing crop recorded on 95 DAS (Table 3). Grass production was lowest on the later sampling dates of 2002.

In the DAS \times treatment interaction for grass yield, the greatest production occurred on 95 DAS in response to 90 kg ha⁻¹ N, and grass production was greatest in response to high N treatment across all DAS (Fig. 1A). The second-highest yields of grass standing crop across DAS were produced by the 45 kg ha⁻¹ N treatment. In contrast, plots with legumes produced slightly less grass than the unfertilized plots. Overall, bermudagrass yield response to level of N fertilizer displayed a slight quadratic increase. Rates of accumulation of grass standing crop differed among treatments during the 50-d study period; 123, 90, and 61 kg ha⁻¹ d⁻¹ were accumulated under 90, 45, and 0 kg ha⁻¹ N, and 53 and 48 kg ha⁻¹ d⁻¹ under lentil and grass pea, respectively. Lower grass production on the legume-treated plots could be attributed to reduced growth of bermudagrass caused by shading or competition for soil moisture from the legumes (Evers, 1985, 2005). Further, fertilizers applied to the legume-treated plots only met the needs of the

Table 2. Mixed model analyses of variance of yield, N concentration, and in vitro digestible dry matter (IVDDM) of grass, legume, and total standing crop in response to treatment (interseeded legume or 0, 45, or 90 kg ha⁻¹ N), days after seeding (DAS), and years.

Source	Grass			Legume			Total Forage		
	Yield	N content	IVDDM	Yield	N content	IVDDM	Yield	N content	IVDDM
DAS	**	**	**	**	**	**	**	**	**
Year	*	**	*	**	**	**	**	**	ns†
Treatment	**	**	**	**	**	**	**	**	**
DAS \times year	**	ns	**	*	ns	ns	**	**	ns
DAS \times treatment	*	**	**	**	**	**	**	**	**
Year \times treatment	ns	ns	ns	ns	ns	ns	ns	ns	ns

* $P < 0.05$.

** $P < 0.01$.

† ns = not significant, $P > 0.05$.

Table 3. Days after seeding (DAS) by year interactions in yield of total standing crop produced by three forage classes on bermudagrass plots, averaged across treatments.

DAS	Forage classes								
	Grass			Legume			Total		
	2001	2002	2003	2001	2002	2003	2001	2002	2003
	kg ha^{-1}								
45	37k†	833j	919j	59h	267g	317g	96j	1100h	1236h
55	2482ef	1571gh	1489h	356fg	455ef	544de	2838f	2026g	2033g
65	3744c	2142fg	2077fg	639d	580d	1753b	4383d	2704f	3830e
75	4210c	3087d	4054c	1337c	1389c	2614a	5547c	4476d	6668b
95	6056a	3243de	4765b	1439c	1289c	2560a	7495a	4532d	7325a

† Values with the same letter within the same forage class were not significantly different ($P > 0.05$).

legumes, so available N to support grass production was in short supply.

Legumes produced more forage in 2003, though there were similarities in amounts among years during the first two DAS in 2002 and 2003, and the last three dates of 2001 and 2002 (Table 3). In the DAS \times treatment interaction in legume standing crop, grass pea produced more ($P < 0.05$) forage than lentil during the last three sampling dates, with roughly three times the biomass on 75 and 95 DAS (Fig. 2B). Rao et al. (2005) reported that grass pea produced three times more biomass than lentils at 75 DAS (5800 vs. 2000 kg ha^{-1} , respectively) when grown in monocultures in cultivated soil during the same growing seasons. Compared with those results, competition from bermudagrass apparently limited legume production in the present experiment. Production of legume standing crop was also likely limited by the

drought conditions during the study. However, the generally small but timely amounts of rain that occurred in each growing season helped support legume growth at critical times in plant development. The higher production by grass pea, relative to lentil, may be related to higher water use efficiency of grass pea, as reported in southern Canada (Biederbeck and Bouman, 1994). Under more typical rainfall regimes for the southern Great Plains and the study site, both legumes would be expected to produce more forage than recorded here.

The DAS \times year interaction in total standing crop was related to higher levels of production across dates in 2003, and similarities in production on 45 and 55 DAS of 2002, and 95 DAS of 2001 and 2003 (Table 3). In the DAS \times treatment interaction in total standing crop, the

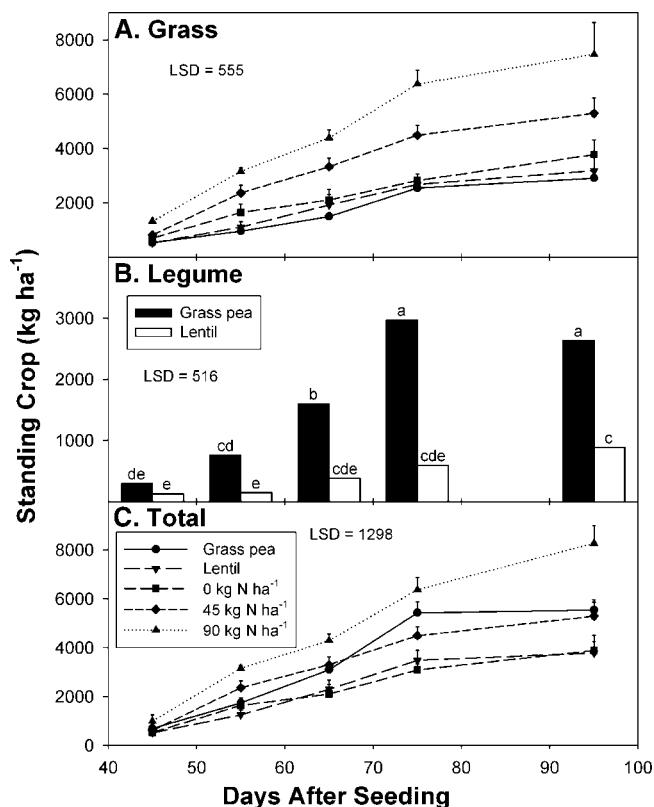


Fig. 1. Days after seeding by treatment interactions in (A) grass, (B) legume, and (C) total standing crop produced by bermudagrass plots; vertical bars noted at points represent one standard error.

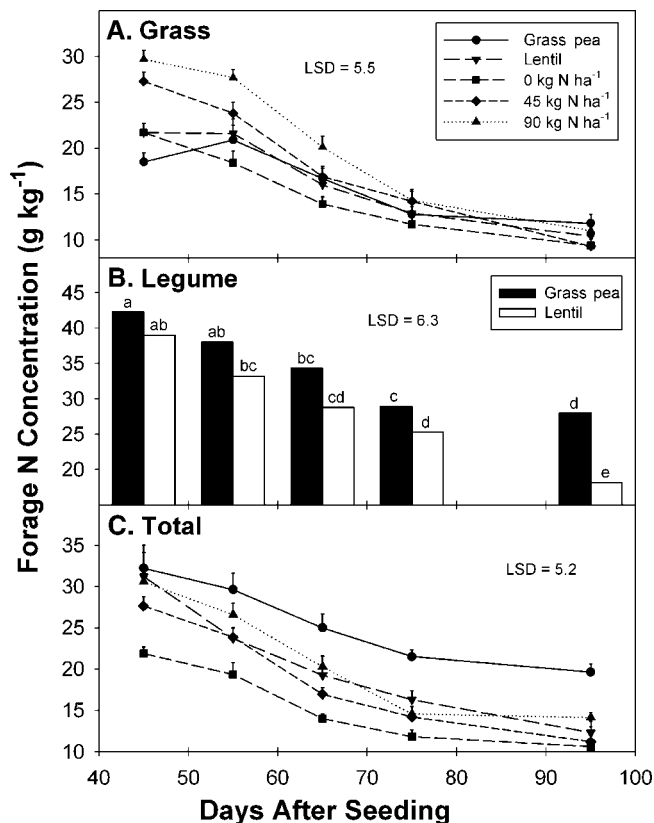


Fig. 2. Days after seeding by treatment interactions in N concentration of (A) grass, (B) legume, and (C) total forage produced by bermudagrass plots; vertical bars represent one standard error, and columns with the same letter were not significantly different ($P > 0.05$).

largest level of standing crop occurred on 95 DAS in response to 90 kg ha⁻¹ N, and was consistently greater across DAS (Fig. 1C). The second-highest yields of total standing crop were recorded for the 45 kg ha⁻¹ N and grass pea treatments. In contrast, plots interseeded with lentil produced levels of total standing crop similar to the unfertilized plots.

Nitrogen Concentration

A significantly higher N concentration was recorded in grass forage during 2003 than the other 2 yr (Table 4), and similar higher levels were noted in N concentration of legume forage during 2001 and 2002. The DAS × year interaction in N concentration of total forage was related to a high value on 45 DAS of 2002, compared with the other years, and consistently higher levels during 55, 65, and 75 DAS in 2003, compared with the other years (Table 5).

As anticipated, N concentration in both grass and legume forage declined during the sampling period (Fig. 2A, 2B). Across all sampling dates, N concentration of grass forage was greatest in response to the 90 kg ha⁻¹ N treatment, followed by grass forage from plots treated with 45 kg ha⁻¹ N through 65 DAS (Fig. 2A). Nitrogen concentrations of grass forage in response to fertilizer treatments were essentially similar during the last two sampling dates. The N concentration of grass standing crop collected from plots receiving legume treatments was generally between levels for forage from the 0 and 45 kg ha⁻¹ N treated plots, with the exception of 45 and 95 DAS. These results suggest some fixed N from the legumes may have transferred to the bermudagrass during the course of the experiment. Legume standing crop in grass pea plots had higher N concentrations than legume standing crop in plots containing lentil (Fig. 2B). The most noticeable differences between grass pea and lentil forage in the DAS × treatment interaction occurred on 75 and 95 DAS. The N concentration was greatest in the plots receiving the grass pea treatment, across all sampling dates except 45 DAS (Fig. 2C). In contrast, the lentil, 45 kg ha⁻¹ N, and 90 kg ha⁻¹ N treatments were similar on most dates, and higher than values recorded for forage from the 0 kg ha⁻¹ N treated plots.

IVDDM Concentration

Year main effects noted in digestibility of legume forage showed higher IVDDM concentrations in 2001

Table 4. Year main effects in variables related to grass and legume forage collected on bermudagrass plots, averaged across treatments.

Years	Grass N	Legume	
		N	IVDDM†
		g kg ⁻¹	
2001	17.0ab‡	29.6b	767a
2002	16.9b	32.1a	744b
2003	18.1a	33.1a	764ab

† IVDDM, in vitro digestible dry matter.

‡ Values with the same letter within columns were not significantly different ($P > 0.05$).

Table 5. Days after seeding (DAS) by year interactions in IVDDM (in vitro digestible dry matter) of grass forage, and N concentration of total forage on bermudagrass plots, averaged across treatments.

DAS	Grass IVDDM			Total forage N		
	2001	2002	2003	2001	2002	2003
	g kg ⁻¹					
45	672cd†	702bc	641def	32.1a	27.3b	25.0bc
55	763a	701bc	714b	24.0c	22.9cd	25.9bc
65	670cde	638ef	624f	17.2ef	19.6e	20.1de
75	633f	522g	527g	13.7g	14.3fg	18.6e
95	619f	486h	548g	11.1g	12.0g	13.4g

† Values with the same letter within forage variables were not significantly different ($P > 0.05$).

and 2003 (Table 4). Within the DAS × year interaction in IVDDM of grass standing crop, 2001 generally had higher IVDDM than forage collected during the other years, with the exception of 45 DAS (Table 5). In the DAS × treatment interaction, concentration of IVDDM in the grass and legume forages declined during the 50-d sampling period (Fig. 3A, 3B). Across the first three sampling dates, IVDDM of grass forage was greatest in response to the 90 kg ha⁻¹ N fertilizer treatment, followed by grass from plots treated with 45 kg ha⁻¹ N. Thereafter, treatment responses were similar, with the exception of forage from the 0 kg ha⁻¹ N treated plots on 95 DAS. The IVDDM concentration of grass forage collected from plots interseeded to a legume generally fell between responses on the 0 and 45 kg ha⁻¹ N treated

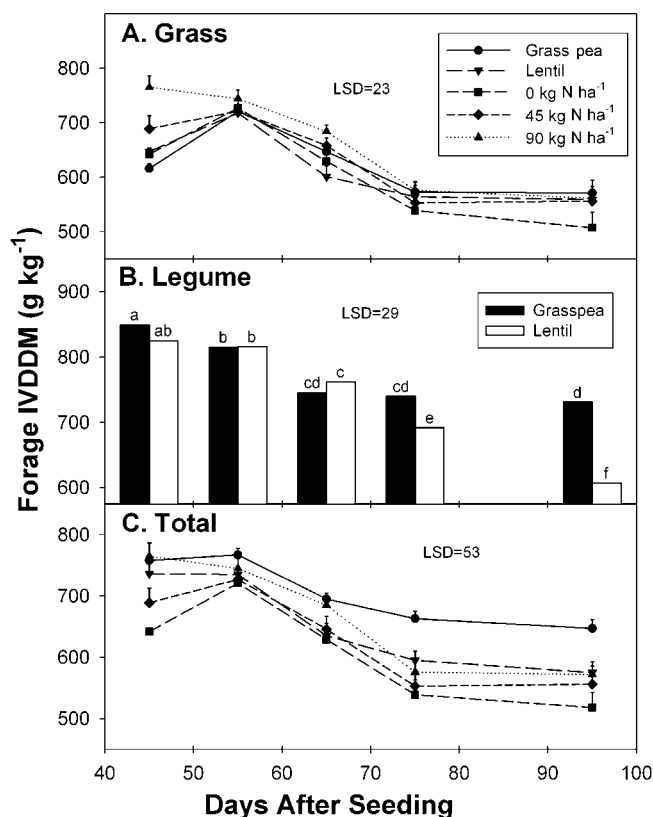


Fig. 3. Days after seeding by treatment interactions in IVDDM (in vitro digestible dry matter) of (A) grass, (B) legume, and (C) total forage produced by bermudagrass plots. Vertical bars noted at points represent one standard error, and columns with the same letter were not significantly different ($P > 0.05$).

plots. The digestibility of forage produced by the legumes on given dates was similar except 75 and 95 DAS, when grass pea forage was more digestible (Fig. 3B). In the DAS \times treatment interaction in IVDDM of total forage, plots seeded to grass pea produced forage with higher digestibility than plots receiving lentil or fertilizer treatments beyond 65 DAS (Fig. 3C). Total forage produced on plots seeded to lentil had IVDDM that was either intermediate to the two higher N treatments, or similar to the 90 kg ha⁻¹ N fertilization. These results were similar to those reported by Sleugh et al. (2000) for birdsfoot trefoil (*Lotus corniculatus* L.) and intermediate wheatgrass [*Thinopyrum intermedium* (Host.) Barkw. & D.R. Dewey], and kura clover (*Trifolium ambiguum* M. Bieb.) and intermediate wheatgrass mixtures. Both combinations had higher IVDDM at later harvest dates than wheatgrass grown under different fertilizer regimes.

Implications

Lentil and grass pea both appear to have some degree of adaptation to the southern Great Plains, and could produce early spring forage to help fill the forage gap that occurs at the end of the wheat graze-out period in May (Redmon et al., 1995; Peel, 2003). However, their value will depend on the grazing system being used, and the forage needed to meet the requirements of the system (Northup et al., 2005). Despite three consecutive years of below-normal precipitation and the competitive effects of the bermudagrass, both legumes produced some forage. The greater productivity of grass pea may be partly due to better water use efficiency, which was 18% greater than lentil in southern Canada (Biederbeck and Bouman, 1994).

A comparison of yield related to treatment-level responses demonstrates that the legumes did not generate improvements in spring standing crop (Fig. 4A). Both lentil and grass pea produced amounts of total forage that were equivalent to the 0 kg ha⁻¹ N and 45 kg ha⁻¹ N treatments. In most instances, the legumes produced <750 kg ha⁻¹ N by mid-May, when bermudagrass began rapid growth. However, neither legume negatively affected production by the bermudagrass. The reduced grass production in the interseeded plots was not entirely due to competition for resources from the legumes. Rather, the limited grass (and hence total) production had some relation to fertilizer management. The lentil and grass pea treated plots were fertilized to meet the needs of the legumes only, so requirements for optimal production by the bermudagrass were not met. Cost-effective increases in production by bermudagrass swards in eastern Oklahoma have been reported in response to 112 to 168 kg ha⁻¹ N in single or multiple applications (Redmon and Woods, 1995; Lalman et al., 2000). The highest fertilizer rate used in our study was lower than those levels, because the lower amounts of precipitation received in central and western Oklahoma limit responsiveness to fertilizer. It should be possible to improve overall production of bermudagrass stands interseeded with annual cool-season legumes by devel-

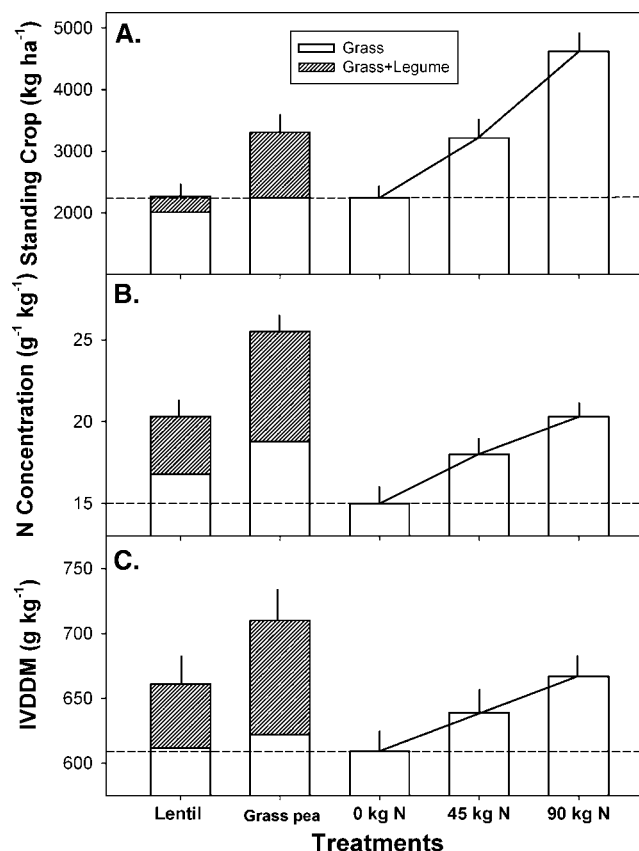


Fig. 4. Mean treatment responses of (A) standing crop, (B) N concentration, and (C) in vitro digestible dry matter (IVDDM) of forage produced by bermudagrass plots to treatments; vertical bars represent one standard error and dashed horizontal lines are effects of the control treatment (0 kg ha⁻¹ N).

oping optimal fertilizer combinations that cost-effectively meet the requirements of both companion crops (Baylor, 1974; Evers, 2005). If fertilizer rates that partially met the spring needs of bermudagrass (for example 45 kg ha⁻¹ N) were combined with enough P to meet requirements for the legumes, levels of standing crop similar to the 90 kg ha⁻¹ N treatment might be possible, along with the concomitant high forage quality associated with the interseeded legumes. Future research is required to develop such guidelines.

Overman et al. (1990) noted substantial increases in dead stems and leaves within bermudagrass swards in the southeastern USA with length of harvest interval, regardless of fertilizer rate. The result of such increases in dead tissues with time is large declines in forage quality, which limits livestock response. Therefore, the impact of legumes on N content and digestibility of total forage is an important factor for interseeding annual legumes into warm-season pasture. Rao et al. (2005) reported up to 168 and 53 kg ha⁻¹ N was stored in aboveground biomass of grass pea and lentil, respectively, when they were grown as monocultures in Oklahoma. Biederbeck et al. (1993) found similar results in Canada, and reported that grass pea and related *Lathyrus* spp. generally retained < 12% of total stored N in the roots and nodules. In this study, grass pea and lentil averaged 41.8 and 8.4 kg ha⁻¹ N, respectively,

stored in aboveground forage at the end of the growing season. Though total production was not improved compared with the fertilizer treatments, the legumes increased both N and IVDDM concentrations in total forage (Fig. 4B, C). Further, small improvements were noted in N concentrations of the bermudagrass. Based on minimum daily forage intakes, mature 454-kg beef cows with suckling calves require roughly 15 g N kg^{-1} of consumed forage (11.5 kg d^{-1}) to meet daily maintenance requirements (National Research Council, 1995). The addition of grass pea to bermudagrass in this study improved N concentrations, to where they exceeded basal requirements during the entire study period, compared with the fertilizer treatments. Nitrogen concentrations of forage from the interseeded plots could also support average daily gains by yearling stocker cattle at levels exceeding or similar to the highest level of N fertilization used in this study. Improving forage quality while sacrificing some production presents an important compromise when planning to interseed legumes into Bermudagrass in the southern Great Plains. Despite the reduction in standing crop, paddocks planted with lentil or grass pea could be used as green supplements for stocker calf production, or to help fill the spring forage deficit period that exists in current forage production systems (Northup et al., 2005).

CONCLUSIONS

In contrast to standing crop, seeding either lentils or grass pea into bermudagrass resulted in large improvements of quality of total forage, compared with fertilizer treatments. Interseeding legumes also improved the N concentration of the grass component, though only marginally, indicating that N stored in roots and nodules of the legumes was being recycled and captured. Seeding grass pea into bermudagrass pasture allowed the quality of forage to exceed that of plant materials produced under the highest N fertilizer rate, while interseeding lentils produced forage quality similar to the highest rate. Under the system of fertilizer management used in this study, interseeding such cool-season annual legumes into bermudagrass may result in a trade-off between forage quality and quantity. Interseeding grass pea into bermudagrass pasture can produce sufficient forage in the spring to allow the initiation of grazing 1 mo earlier, and significantly improve forage quality through midsummer.

ACKNOWLEDGMENTS

We wish to acknowledge Dakota Frontier Seeds, Ltd., for supplying AC Greenfix seed, and Delmer Shantz for providing technical and field assistance.

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